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Phase II Pilot Study of Vemurafenib in Patients With Metastatic *BRAF*-Mutated Colorectal Cancer

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See accompanying editorial on page 3990 and article on page 4023

A B S T R A C T

Purpose

*BRAF*V600E mutation is seen in 5% to 8% of patients with metastatic colorectal cancer (CRC) and is associated with poor prognosis. Vemurafenib, an oral *BRAF* V600 inhibitor, has pronounced activity in patients with metastatic melanoma, but its activity in patients with *BRAF* V600E-positive metastatic CRC was unknown.

Patients and Methods

In this multi-institutional, open-label study, patients with metastatic CRC with *BRAF* V600 mutations were recruited to an expansion cohort at the previously determined maximum-tolerated dose of 960 mg orally twice a day.

Results

Twenty-one patients were enrolled, of whom 20 had received at least one prior metastatic chemotherapy regimen. Grade 3 toxicities included keratoacanthomas, rash, fatigue, and arthralgia. Of the 21 patients treated, one patient had a confirmed partial response (5%; 95% CI, 1% to 24%) and seven other patients had stable disease by RECIST criteria. Median progression-free survival was 2.1 months. Patterns of concurrent mutations, microsatellite instability status, CpG island methylation status, PTEN loss, EGFR expression, and copy number alterations were not associated with clinical benefit. In contrast to prior expectations, concurrent *KRAS* and *NRAS* mutations were detected at low allele frequency in a subset of the patients' tumors (median, 0.21% allele frequency) and were apparent mechanisms of acquired resistance in vemurafenib-sensitive patient-derived xenograft models.

Conclusion

In marked contrast to the results seen in patients with *BRAF* V600E-mutant melanoma, single-agent vemurafenib did not show meaningful clinical activity in patients with *BRAF* V600E mutant CRC. Combination strategies are now under development and may be informed by the presence of intratumor heterogeneity of *KRAS* and *NRAS* mutations.

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INTRODUCTION

BRAF mutations are present in between 5% and 15% of colorectal cancer (CRC). Classically, these mutations are common in the sessile serrated adenoma pathway of adenoma to carcinoma progression and are associated with unique molecular features, including microsatellite instability, hypermethylation, and minimal chromosomal instability.¹⁻³ *BRAF* mutations are present at higher frequency in right-sided tumors and are more likely to present with poorly differentiated and node-positive tumors. *BRAF* mutations frequently coexist with high microsatellite instability and provide

modest prognostic information for recurrence.^{4,5} Patients with metastatic *BRAF*-mutated CRC have a poor prognosis, with a median survival of approximately 12 months despite the utilization of combination chemotherapy.⁶

The mutation of a valine to glutamate in codon 600 in the *BRAF* kinase domain (V600E) results in a constitutively active protein. The V600E mutation accounts for approximately 95% of the activating mutations in *BRAF* in CRC. This mutation results in activation of the MAPK pathway, as highlighted by the near mutual exclusivity with activating mutations in *KRAS* and *NRAS*. The unique conformation induced by V600E mutations is inhibited by

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vemurafenib, a kinase inhibitor designed to have increased affinity for the mutated form of *BRAF*. Treatment with vemurafenib has demonstrated a high response rate and prolonged progression-free survival in *BRAF* mutant melanoma, leading to US Food and Drug Administration approval for this indication.⁷ Preclinical studies in *BRAF* mutant CRC cell line models suggested activity with vemurafenib, with growth arrest and inhibition of MAPK signaling, and provided a strong rationale for clinical evaluation.⁸

However, the activity of vemurafenib in patients with CRC with *BRAF* V600 mutations had not been defined. Therefore, we conducted a multisite expansion cohort on the phase I trial of vemurafenib after establishment of the recommended phase II dose in melanoma.⁷ To explore the biology and potential predictive markers of response to therapy, we subjected patient material for correlative studies based on the emerging data on the distinct biology of *BRAF* mutant CRC.

PATIENTS AND METHODS

Eligibility Criteria

For the CRC expansion cohort, eligible patients had metastatic CRC with centrally confirmed V600E-positive *BRAF*, evaluable or measurable disease by RECIST version 1.0, and Eastern Cooperative Oncology Group performance status of 0 or 1. Patients also had to be ≥ 18 years old with adequate hematopoietic, hepatic, and kidney function and a life expectancy ≥ 3 months. Key exclusion criteria included chemotherapy within 2 weeks, prior therapy with targeted agents against *BRAF* or *MEK*, uncontrolled CNS metastases, and mean QTc ≥ 500 milliseconds. Given the risk of squamous cell carcinoma (SCC) with *BRAF* inhibition, patients were required to be free of pre-existing nonmelanoma skin lesions including keratoacanthoma, actinic keratosis, and squamous carcinoma in situ. The protocol (ClinicalTrials.gov identifier: NCT00405587) was approved by the institutional review boards of all participating institutions, and written informed consent was obtained for all patients before performing study-related procedures.

Drug Administration and Study Design

Vemurafenib was provided in microprecipitated bulk powder formulation as 240-mg film-coated tablets, dosed at the previously determined maximum-tolerated dose of 960 mg orally twice a day, and administered continuously in 28-day cycles.⁹ Patients underwent history and physical examination, CBC, chemistries, coagulation parameters, urinalysis, carcinoembryonic antigen, ECG, and tumor measurements within 3 weeks before start of therapy. While on study, patients underwent evaluations, including brief history and physical examination, vital signs, urinalysis, CBC, and chemistries, on days 8, 15, and 29 during cycle 1 and on day 1 of each subsequent cycle. ECGs were performed at baseline and before cycle 2. Adverse events were classified according to the Common Terminology Criteria of Adverse Events (version 3.0). Response was assessed radiographically after every two cycles or more frequently at the treating physician's discretion. Given the risk of SCC with *BRAF* inhibition, dermatologic, head, neck, and chest (with chest computed tomography) monitoring for SCC were performed at baseline and periodically thereafter.

Pharmacokinetic Studies

Pharmacokinetic samples were obtained on days 1 and 15 (before dose and 0.5, 1, 2, 4, and 8 hours after dose), day 16 (before dose), and day 29 (before dose). During continued dosing, pharmacokinetic samples were obtained before dose once every 4 weeks. Vemurafenib plasma concentration was evaluated by noncompartmental methods. Pharmacokinetic samples were also collected from the parallel expansion cohort in melanoma, as previously reported.⁹

Correlative Studies

DNA was extracted from macrodissected tumor on formalin-fixed paraffin-embedded (FFPE) slides, including hematoxylin and eosin-stained reference slide according to manufacturer methods (Qiagen DNeasy, Qiagen, Hilden, Germany; and Epicentre, Madison, WI).¹⁰ To determine copy number and hotspot gene mutations, the tumor was subjected to OncoScan FFPE Express V2 (Affymetrix, Santa Clara, CA) and Sequenom MassARRAY (Sequenom, San Diego, CA), both of which were optimized for use of FFPE samples with a sensitivity of 10% mutant alleles. The mutations detected are included in hotspot regions in *KRAS*, *NRAS*, *PIK3CA*, and *AKT*, among others (Data Supplement). Copy number alterations were determined by segmentation analysis produced by Nexus Copy Number (BioDiscovery, El Segundo, CA). PTEN and EGFR protein expression was determined using immunohistochemistry as previously described (PTEN clone 6H2.1, Dako, Carpinteria, CA; EGFR clone 31G7, Zymed, San Francisco, CA).^{11,12} Methylation analysis was performed to classify patients based on the degree of tumor methylation using bisulfite polymerase chain reaction (PCR) followed by pyrosequencing for a previously established six-marker CpG island methylator phenotype (CIMP) panel (MINT1, MINT2, MINT31, hMLH1, p16, and p14).^{13,14}

To assess low-frequency clones, DNA was subjected to emulsion PCR (BEAMing, Sysmex Inostics, Baltimore, MD), a methodology to perform single-molecule PCRs on magnetic beads in water-in-oil emulsions, followed by allele-specific hybridization and flow cytometry-based allele quantification with sensitivity of 0.15% on FFPE tissue specimens (Data Supplement).¹⁵ A complementary droplet digital PCR was applied to a second cohort of 19 *BRAF* mutant tumors from patients not treated on the protocol but obtained from the Royal Melbourne Hospital (Parkville, Victoria, Australia). Analyses were performed with a droplet digital PCR *KRAS* screening multiplex kit (BioRad QX100, BioRad, Hercules, CA) according to the manufacturer's instructions, with sensitivity to 0.15% mutant allele frequency for FFPE samples and 0.05% for fresh tissue. Patient-derived xenografts were derived from core biopsies and implanted in NOD-SCID- γ mice (Jackson Labs, Bar Harbor, ME). Established tumors were treated with vemurafenib analog PLX4720. Tumor sequencing of the resistant xenograft tumors for 409 exons was performed by Ion AmpliSeq Comprehensive Cancer Panel on the Ion Torrent PGM (Life Technologies, Carlsbad, CA).

RESULTS

Patient Characteristics and Treatment

Twenty-one patients with *BRAF* V600E mutation-positive metastatic CRC (20 colon cancers and one rectal cancer) were enrolled at multiple institutions in the United States and Australia. Of the 21 patients, 11 were men and 10 were women, with a median age of 65 years (range, 38 to 91 years; Table 1). Baseline Eastern Cooperative Oncology Group performance status was 0 and 1 in 11 and 10 patients, respectively. Results from local standard *KRAS* testing (codons 12 and 13) were available for 16 of the 21 patients, with all tumors characterized as wild type. All patients received prior therapy, and all except one patient received at least one prior chemotherapy regimen in the metastatic setting; 14 patients received at least two lines. Consistent with the known chemotherapy-refractory nature of the disease, only three of the 20 previously treated patients achieved a partial response to prior chemotherapy treatments.¹⁶

Safety

The most common toxicities of any grade while on treatment were fatigue, hyperglycemia, proteinuria, and diarrhea (Table 2). There were no grade 4 toxicities, and the most common grade 3 toxicity was SCC of the skin (a Common Terminology Criteria for Adverse Events term that was inclusive of keratoacanthomas). Overall, 15 of 21 patients experienced at least one grade 3 AE while on study,

Table 1. Baseline Demographic and Clinical Characteristics

Characteristic	No. of Patients	%
Sex		
Male	11	52
Female	10	48
Age, years		
Median	65	
Range	38-91	
ECOG performance status		
0	11	52
1	10	48
Race		
White	19	91
Asian	2	9
Previous therapies		
Surgery	21	100
Chemotherapy	20	95
Radiation	2	10
No. of previous therapies for metastatic disease		
0	1	5
1	1	5
2	4	19
≥ 3	15	71

Abbreviation: ECOG, Eastern Cooperative Oncology Group.

with seven patients experiencing severe adverse events attributed to the therapy. There were no toxicity-related discontinuations or deaths, but 52% of patients (11 patients) had AEs leading to dose interruption across all cycles. The most common causes of dose interruption included rash (n = 4), elevated liver function tests (n = 2), and diarrhea (n = 2). Furthermore, 19% of patients (four patients) required one or more dose reductions secondary to drug-related toxicities of rash, fatigue, nausea, or anorexia.

Table 2. Summary of All Adverse Events While Receiving Treatment in ≥ 25% of Patients, Irrespective of Causality

Adverse Event	No. of Patients (%)	
	Grade ≥ 3	All Grades
Squamous cell carcinoma of skin*	5 (23.8)	5 (23.8)
Hyperbilirubinemia	3 (14.3)	7 (33.3)
Rash	2 (9.5)	7 (33.3)
Hyponatremia	2 (9.5)	6 (28.8)
Fatigue	1 (4.8)	13 (61.9)
Diarrhea	1 (4.8)	8 (38.1)
Arthralgia	1 (4.8)	8 (38.1)
Vomiting	1 (4.8)	7 (33.3)
Lymphopenia	1 (4.8)	6 (28.8)
Proteinuria	0	9 (42.9)
Photosensitivity reaction	0	7 (33.3)
Hyperglycemia	0	7 (33.3)
Nausea	0	7 (33.3)
Hypalbuminemia	0	6 (28.6)
Pyrexia	0	6 (28.6)
Headache	0	6 (28.6)

*Cutaneous squamous cell carcinomas were reported as grade 3 adverse events, and a majority were managed by excision. One patient required treatment discontinuation for numerous lesions unable to be adequately managed by excision.

Patients were monitored closely and underwent scheduled skin exams by dermatologists during therapy. Five patients developed new cutaneous SCCs while on treatment (Appendix Fig A1, online only). In addition, two patients developed seborrheic dermatitis, and one patient each developed a skin papilloma and actinic keratosis. There were no SCCs at other monitored sites including the head, neck, and lung. Other cutaneous adverse events included photosensitivity in 33.3% (seven patients), rash in 19% (four patients), dry skin in 24% (five patients), pruritus in 14% (three patients), and hyperkeratosis in 10% (two patients).

Given the reported potential for vemurafenib to induce QT prolongation, ECGs were evaluated at baseline and the beginning of cycle

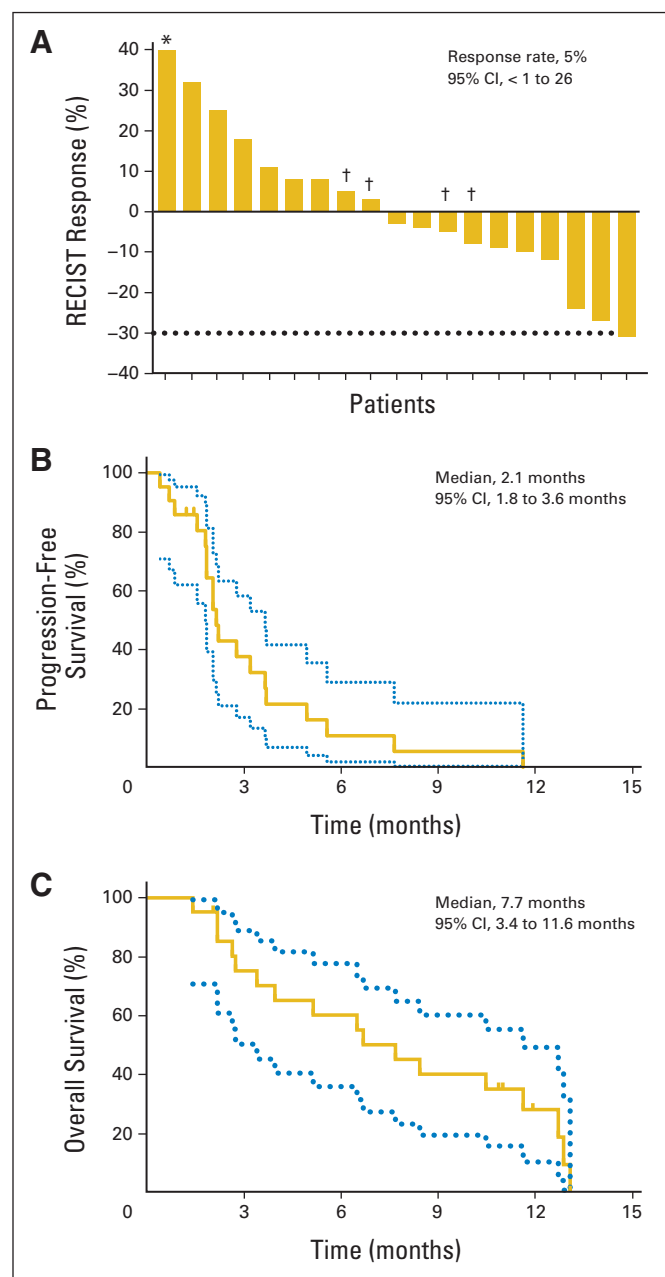


Fig 1. (A) Waterfall plot of best RECIST response to vemurafenib in evaluable patients. (*) Fifty-percent growth from baseline. (#) Patients with notable mixed responses. (B) Progression-free survival. (C) Overall survival.

2, with a mean increase in QTc of 12.5 milliseconds (standard deviation, 17.8 milliseconds). Three patients experienced an increase greater than 30 milliseconds and less than 45 milliseconds.

Pharmacokinetic studies were available on all patients. When compared with melanoma patients in the concurrently enrolled cohort, there was no statistically significant difference in vemurafenib concentration after the first dose (mean $\pm$ standard deviation: maximum concentration, $5.5 \pm 2.3 \mu\text{g/mL}$; area under the curve from 0 to 8 hours, $31.5 \pm 15.1 \mu\text{g/mL}\cdot\text{h}$; [Appendix Fig A2](#), online only). Thirteen patients were available for day 15 sampling, with a maximum concentration of $38.6 \pm 15.9 \mu\text{g/mL}$ and area under the curve from 0 to 8 hours of $262.4 \pm 101.4 \mu\text{g/mL}\cdot\text{h}$.

Antitumor Activity

Nineteen of the 21 patients in this trial had postbaseline scans. There were no complete responses. One patient had a confirmed partial response, which was durable for 21 weeks. Seven other patients had stable disease as best response for at least 8 weeks (range, 8 to 50 weeks; [Fig 2A](#)). Ten patients had some degree of tumor shrinkage on the first planned computed tomography scan. Three patients had pretreatment and follow-up [^{18}F]fluorodeoxyglucose positron emission tomography; two of these patients had a metabolic response with greater than 25% reduction in standard uptake value ([Appendix Fig A3](#), online only). The median progression-free survival was 2.1

months (range, 0.4 to 11.6 months), with two patients free from progression for more than 6 months. Median overall survival time was 7.7 months (range, 1.4 to 13.1 months).

Correlative Biomarkers

To evaluate the lower than expected overall response rate (compared with that seen in melanoma) and heterogeneous response to therapy in some patients, baseline tumor tissue from requested archival FFPE slides was analyzed for molecular and protein biomarkers in exploratory analyses. Prior preclinical studies had suggested that activation of the PI3K pathway may be associated with resistance to *BRAF* inhibition based on correlation to sensitivity in a panel of *BRAF* mutant CRC cell lines.^{8,17} The PI3K pathway was activated by complete loss of PTEN protein staining or mutations in *PIK3CA* or *AKT* in four of 14 evaluable tumors, with no association with response to therapy ([Appendix Fig A4](#), online only). Because EGFR has been implicated in feedback activation of the MAPK pathway after *BRAF* inhibition, total expression of EGFR in the tumor was evaluated as a potential predictive biomarker. When dichotomized based on any expression versus no expression, there was no correlation with tumor response ([Appendix Fig A4B](#)).

Hypermethylation is a nearly universal feature of tumors with *BRAF* mutations, and analysis of a previously established six-gene CIMP panel confirmed this finding.¹⁸ The pattern of methylation in

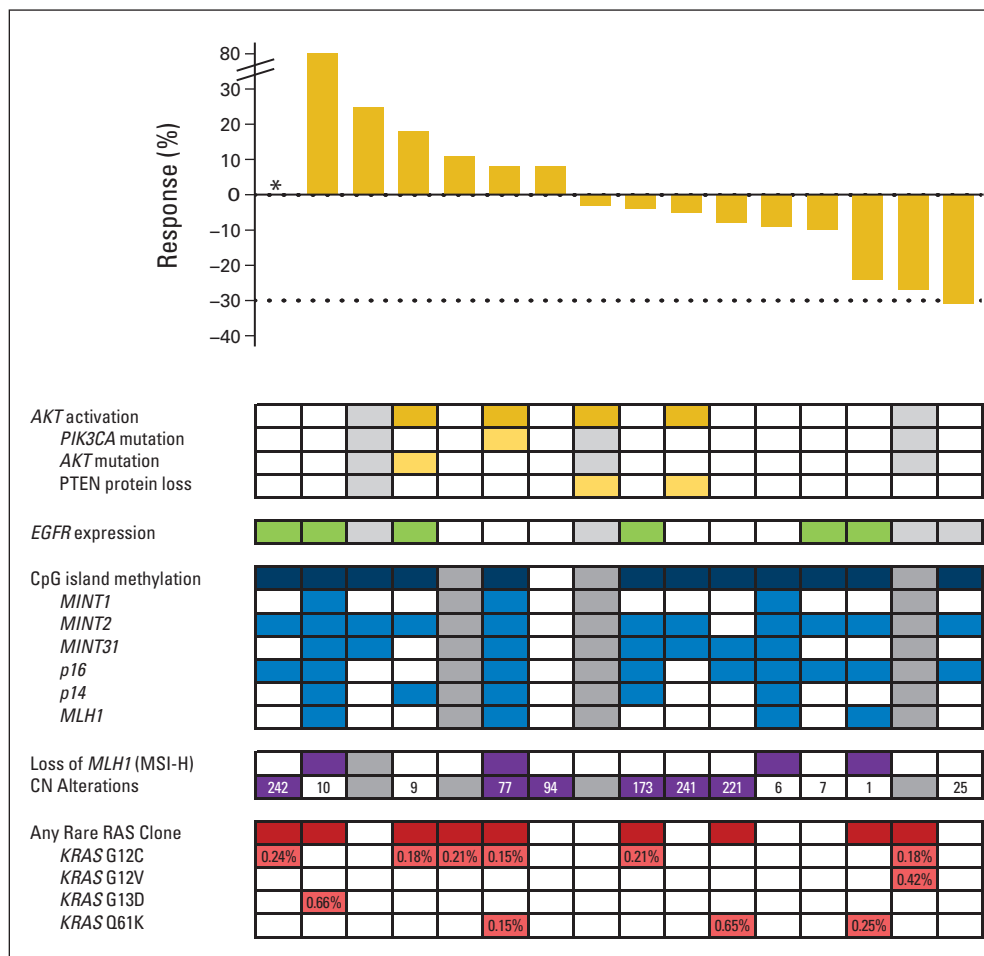


Fig 2. Correlation of best overall response rate with molecular characterization. Gray boxes represent samples unable to be analyzed for the specified assay. Numbers in the copy number (CN) alteration boxes represent the number of unique segments with CN alterations. Percentages in the RAS clone box represent mutant allele frequency. (*) Not evaluable for response.

these markers has been previously used to subclassify CIMP-high tumors but did not correlate with clinical activity in this cohort. Complete loss of hMLH1 by protein expression was seen in patients with methylation of hMLH1 in the promoter region and defined a subset of patients (four of 15 patients) with a high level of microsatellite instability. Tumors were further annotated by the number of unique segments with copy number alterations. The expected inverse correlation between higher levels of copy number alteration and microsatellite instability was seen, but neither correlated with radiographic response.

Prior studies have demonstrated the presence of rare *KRAS* or *NRAS* clones that can lead to resistance in patients treated with EGFR monoclonal antibodies. To explore whether a similar finding is seen in *BRAF*-mutated CRC, tumors were subjected to a high-sensitivity emulsion PCR methodology to evaluate for the presence of rare *KRAS* or *NRAS* mutant clones in the pretreatment tissue. Low-frequency *KRAS* or *NRAS* mutations were seen in nine of 16 evaluable tumors (56%), with a median allele frequency of 0.21% (interquartile range, 0.18% to 0.42%; Fig 3A). These mutations were not detected by standard-of-care clinical sequencing or mass spectroscopy genotyping with a sensitivity of 10%. There was no correlation of the presence of

low-frequency alleles with radiographic response or duration of treatment. To confirm this finding, a second cohort of primary *BRAF* V600E mutant CRC tumors was evaluated for low-frequency *KRAS* mutations by an independent digital PCR methodology. Thirteen (68%) of the 19 tumors evaluated demonstrated detectable *KRAS* mutations at a median allele frequency of 0.31% (interquartile range, 0.24% to 0.61%). Postprogression tissue was not available from patients who initially developed a response to directly determine the clinical relevance of these low-frequency mutations. Instead, we treated a vemurafenib-sensitive *BRAF* V600E patient-derived xenograft with vemurafenib until progression and explored mechanisms of resistance (Fig 3B). In both cases, the implanted tumor had low-frequency *KRAS* clones present at 0.07% and 0.08% by droplet digital PCR. Tumor regression was seen before development of resistance within 8 weeks. Two separate resistant tumors were sequenced, confirming persistent presence of *BRAF* V600E, with *KRAS* G12D or G12R mutations present at an allele frequency of greater than 40%.

DISCUSSION

CRCs with mutations in *BRAF* V600E represent a unique CRC subtype with poor prognosis and limited response to standard-of-care therapies. In contrast to melanoma, the tumors have limited response to single-agent *BRAF* inhibition. This discrepancy is not attributed to differences in dose-limiting adverse events or pharmacokinetics but is tumor specific and suggests either differences in oncogene dependency or pharmacodynamic failure as a result of feedback mechanisms. The presence of the mutation in early sessile serrated adenomas and the ability of the *BRAF* V600E mutation to recapitulate tumor development in a genetically engineered mouse model of CRC suggest that this mutation is a driver mutation important for carcinoma initiation.^{19,20} The population of enrolled patients with *BRAF* mutant CRC reflected the anticipated molecular phenotype, with high levels of DNA methylation, few copy number alterations, and enrichment for microsatellite instability. Our results demonstrate no meaningful differences in pharmacokinetics between patients with CRC and melanoma, with similar adverse event profiles and administered dose-intensity.²¹ Although vemurafenib is a substrate for cellular drug exporters such as P-glycoprotein, which is upregulated in CRC relative to melanoma, a similar low response rate (one responder) with the non-P-glycoprotein substrate *BRAF* inhibitor dabrafenib has been reported in patients with CRC.²²

Preclinical work has suggested that the MAPK pathway is insufficiently inhibited with single-agent *BRAF* inhibition in CRC, in contrast to what is considered adequate inhibition in melanoma, irrespective of the *BRAF* inhibitor used.^{17,23,24} This alone may be sufficient to limit clinical activity in CRC. In addition, in the clinical setting in melanoma, there seems to be a threshold of inhibition as measured by phospho-Erk expression required to induce response.²⁵ Because this study did not incorporate on-study biopsies, direct assessment of the ability of vemurafenib to sufficiently inhibit the MAPK pathway is not possible. Preclinical studies since this clinical trial was first reported have added considerably to our understanding of MAPK inhibition and early reactivation of the pathway in CRC, with perhaps the most compelling reactivation being through feedback EGFR reactivation.^{23,24} It has been proposed that EGFR expression levels may predict for this reactivation; however, no correlation was seen between

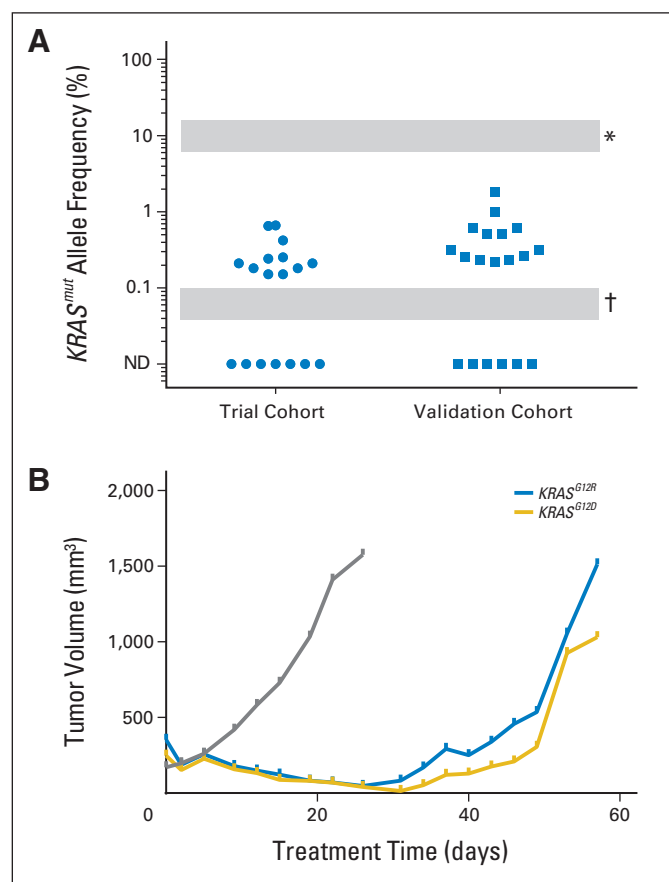


Fig 3. (A) Rare *KRAS*-mutant (mut) clones were detected in a validated cohort of *BRAF* V600E-mutant colorectal cancer tumors by droplet digital polymerase chain reaction (ddPCR). (*) Lower limit of detection for clinical sequencing is approximately 5% to 20% depending on assays used. (†) Lower limit of detection for ddPCR. (B) A patient-derived xenograft model developed resistance to vemurafenib analog PLX4720 provided through ad lib chow at a dose of 417 mg/kg. Gray represents control arm; blue and gold represent PLX4720 treatment arm with acquisition of *KRAS* G12R and G12D mutations, respectively.

total EGFR expression in the primary tumor and clinical activity in our study, suggesting that baseline EGFR expression may be a poor correlate of subsequent EGFR pathway reactivation after *BRAF* inhibition. The combination of EGFR and vemurafenib is being explored in several clinical trials, with some promising early results with panitumumab and an ongoing randomized study of cetuximab and irinotecan, with or without vemurafenib (ClinicalTrials.gov identifier: NCT02164916).²⁶ In contrast, prior preclinical studies suggested that PI3K pathway activation may be associated with resistance to *BRAF* inhibition. However, neither PTEN protein loss nor *PIK3CA* mutation was correlated with resistance in our study, consistent with the limited additional clinical activity seen with the addition of a PI3K inhibitor to *BRAF* and EGFR inhibition.²⁷

The finding of *KRAS* and *NRAS* mutations with low allele frequency in the *BRAF* mutant tumors is unexpected. With rare exception, *BRAF* V600E and *KRAS*/*NRAS* mutations are mutually exclusive when assessed by standard sensitivity sequencing.²⁸⁻³⁰ Acquisition of *NRAS* mutations has been described as a mechanism of resistance to *BRAF* inhibition in melanoma, and our patient-derived xenograft models suggest that a similar mechanism may be present in *BRAF*-sensitive CRCs.³¹ Because low allele frequency *KRAS*/*NRAS* mutations have been associated with resistance to cetuximab and current clinical trials are combining *BRAF* and EGFR inhibition, there is potential for these low allele frequency *KRAS*/*NRAS* mutations to be important drivers for resistance to the combination. The allele frequency observed in the patient-derived xenograft models is consistent with *BRAF* and *KRAS* mutations coexisting in the same tumor clones, suggesting that strategies to address both mutations will be needed. Evaluation of *KRAS*/*NRAS* mutations should be incorporated into future clinical trials of *BRAF* inhibition, including assessment at the time of progression. Other described mechanisms of acquired resistance in CRC to *BRAF* inhibition in preclinical models and patients include EGFR, *KRAS*, and *MET* amplification.³²

The contrast of activity from *BRAF* inhibition in melanoma and CRC has been an important cautionary tale for the trend toward tumor-agnostic, oncogene-defined basket clinical trials.

Histology can be a surrogate for potentially important differences in biology. *BRAF*-mutated CRC and melanoma differ in the tissue lineage (endoderm v ectoderm, respectively) and concurrent epigenetic features (nearly universal CIMP positive v CIMP negative, respectively), among other factors. *BRAF* inhibition has demonstrated some single-agent activity in non-small-cell lung cancer and thyroid cancer, among others, although the magnitude of the benefit remains to be defined.^{33,34} In each setting, alternate mechanisms of resistance to *BRAF* inhibition have been proposed, reiterating the concept that tumor histology is still a major determinant of sensitivity to targeted therapies. Further studies should leverage these similarities and differences in *BRAF* inhibition sensitivity to optimize future combination strategies.

AUTHORS' DISCLOSURES OF POTENTIAL CONFLICTS OF INTEREST

Disclosures provided by the authors are available with this article at www.jco.org.

AUTHOR CONTRIBUTIONS

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Financial support: Scott Kopetz

Provision of study materials or patients: Scott Kopetz, Jayesh Desai, Emily Chan, Leonard Saltz

Collection and assembly of data: Scott Kopetz, Emily Chan, Joel Randolph Hecht, Peter J. O'Dwyer, Dipen Maru, Van Morris, Arvind Dasari, Woonbook Chung, Peter Gibbs, Brian James, Garth Powis, Keith B. Nolop, Suman Bhattacharya

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Manuscript writing: All authors

Final approval of manuscript: All authors

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AUTHORS' DISCLOSURES OF POTENTIAL CONFLICTS OF INTEREST

Phase II Pilot Study of Vemurafenib in Patients With Metastatic *BRAF*-Mutated Colorectal Cancer

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Appendix



Fig A1. Development of keratoacanthoma while on therapy. (A) Before initiation of therapy and (B) after 8 weeks of treatment. (C and D) Development of multiple keratoacanthomas requiring treatment discontinuation.

Vemurafenib in *BRAF*-Mutated Colorectal Cancer

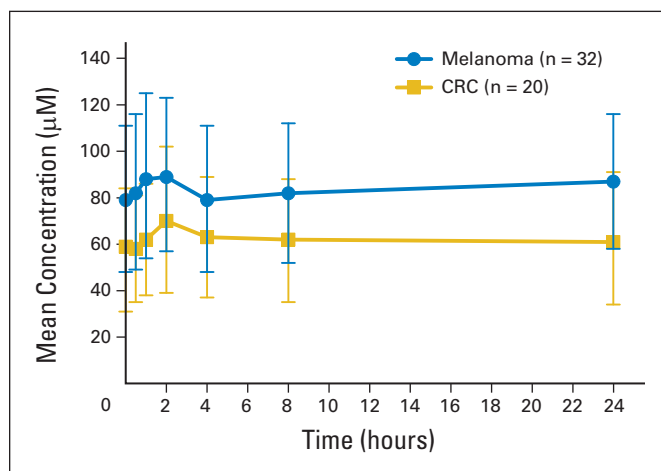


Fig A2. Mean steady-state concentration-time profile of patients with metastatic colorectal cancer (CRC) compared with patients with melanoma treated with vemurafenib 960 mg twice per day.

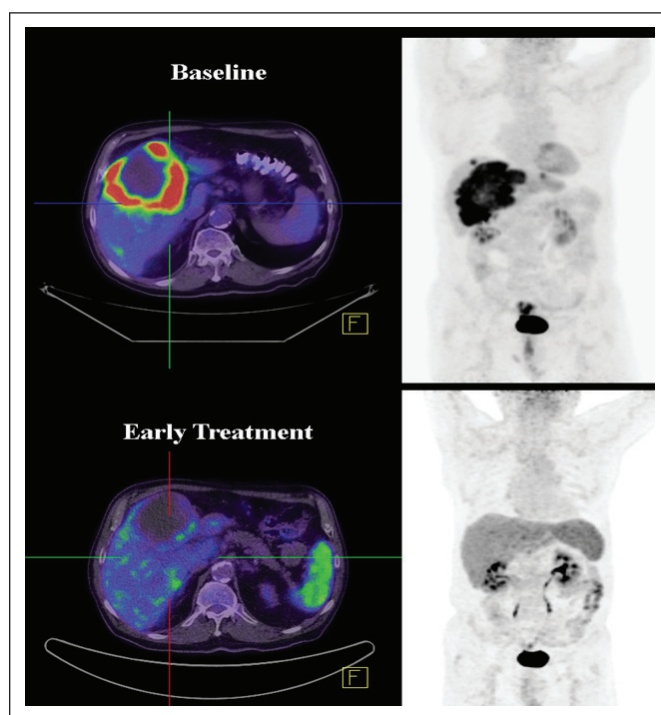


Fig A3. Response by positron emission tomography to vemurafenib. (Top) Baseline scan. (Bottom) After one cycle of vemurafenib.

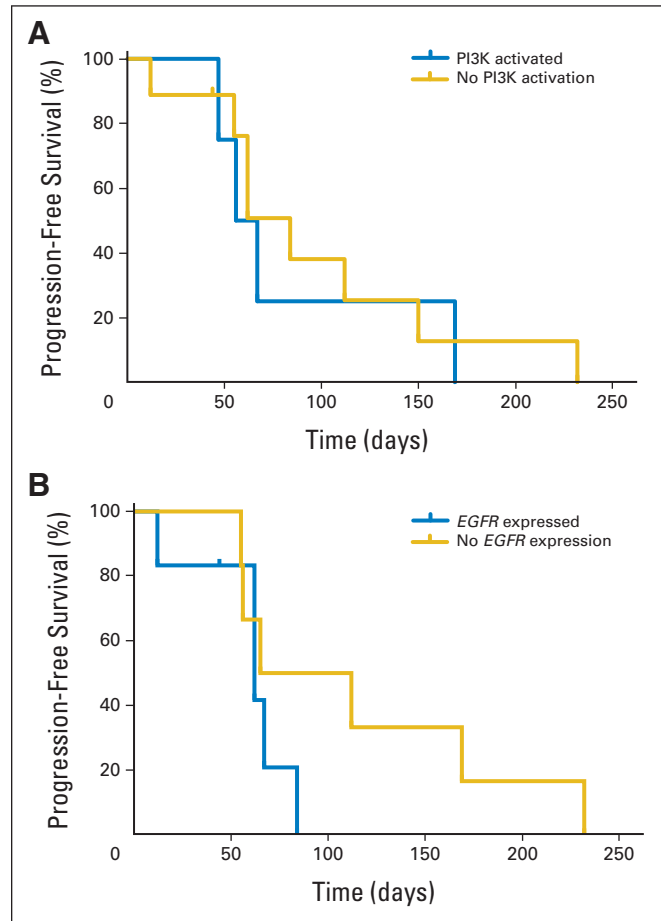


Fig A4. (A) Progression-free survival (PFS) for patients with or without identified alterations resulting in PI3K pathway activation (as denoted in Fig 2). (B) PFS for patients with tumors with intact or absent EGFR expression by immunohistochemistry.